

reagents such as buffers, enzymes, substrates for detection, etc. Once data collection has been performed, a fast buffer flow is introduced, e.g., via the channel which is oriented perpendicular to the long axis of the magnetic islands. This aids in stripping the magnetic particles off the chip. While either channel can be used, stripping may be much more efficient when using a flow direction perpendicular to the trapping field,  $H_x$ . The channels can then be flushed, and the chip is ready for reuse.

**[0138]** Any appropriate fabrication technique may be employed to make the microfluidic assembly. The selection may depend upon the choice of materials. Appropriate techniques include various micromachining and microfabrication techniques, including film deposition processes such as spin coating, chemical vapor deposition, etching techniques, injection molding, etc. Well-known bonding methods may be used to bond a material such as glass to a substrate such as silicon. Straightforward heat bonding, which is compatible with the magnetic chip design and fabrication processes described above, may be used. Another bonding technique is anodic bonding, in which a high strength electric field is used to bond the assembly, reducing the need for high temperatures. According to this technique, a glass sheet (which can be pre-etched with the desired pattern of channels) is placed on a bare or oxidized silicon wafer, which has been patterned with magnetic regions (e.g., prepared up till Step 5 in **FIG. 10**). After proper alignment of the channels with the magnetic islands, the assembly is heated to approximately 350 C. while holding the substrate at a positive (anodic) potential relative to the glass. Voltages on the order of 500 V are typical, though any appropriate voltage may be used. This causes the surfaces at the interface to diffuse to form a permanent bond. After bonding, the magnetic regions are magnetized as described above. The conditions for anodic bonding are compatible with the chip fabrication processes, so modification of those processes should not be required.

**[0139]** As will be appreciated by one of ordinary skill in the art, numerous variations in terms of the design, materials, and fabrication technology for the microfluidic assembly may be made. In addition, devices such as pumps, tubing, heating elements, etc., may be attached to and employed in conjunction with the microfluidic assembly. Devices such as pumps (e.g., electrokinetic pumps), heating elements, etc., may be provided either on-chip or off-chip. In certain embodiments of the invention the materials used in fabrication of the microfluidic components and ancillary equipment is nonmagnetic.

**[0140]** In general, microfluidic systems and related devices and components are well known in the art. Various aspects of these technologies are described, for example, in U.S. Pat. No. 5,603,351 and PCT US/17116 (sample cassettes); WO96/39260 (formation of fluid-tight electrical conduits); U.S. Pat. No. 5,747,169 (sealing), and WO/71243 and references therein for general description.

**[0141]** (2) Integrated Photodetectors

**[0142]** As described below, many of the schemes for detecting interactions between probes and samples and/or for encoding and decoding bead identities rely on optical detection schemes such as fluorescence detection. Thus in certain embodiments of the invention on-chip photodetectors are provided in proximity to the attachment sites for

magnetic particles for detection of signals from beads, probes, and/or targets (e.g., fluorescent or luminescent signals). Photodetector technology on substrates such as silicon is well known (see, e.g., U.S. Pat. No. 5,965,452), and methods for producing integrated photodetectors (e.g., lithographic processes) are compatible with the other fabrication steps for the magnetic chip. The photodetector element (e.g., a charge-coupled device (CCD) structure, MOS photodiode, etc.) may be covered with a transparent material such as glass or plastic for protection.

**[0143]** Various possibilities exist for integrating photodetectors into the magnetic chip of the invention. In one embodiment, referring to **FIG. 12**, integrated photodetectors **35** can be built on a silicon substrate **25** that forms the surface of the chip. Magnetic regions may be formed prior to formation of the photodetectors. Alternatively, the photodetectors may be formed first, and the magnetic regions formed afterwards. The photodetectors can be distributed in a regular pattern that substantially corresponds to the pattern of gaps between magnetic regions, and can be substantially equal in number to the number of magnetic regions. Circuitry (not shown) coupled to the photodetectors can transmit the fluorescent signals to a processor which can process the signals into an image-map which can be analyzed.

**[0144]** Including optical detection capabilities on the chip itself offers a number of advantages. Due to the proximity of the on-chip photodetectors to the arrayed beads, the sensitivity of this detection scheme will likely be significantly superior to confocal scanning. This may be important when detecting genomic targets under conditions in which relatively few target molecules are present, which is likely to be an increasingly important future direction for high-density array technologies. Photodetector integration onto the chip will further enhance the photon capture efficiency. The reusability of the chip over multiple arraying runs may make on-chip detection an economically feasible approach. On-chip detection enhances the portability of the system since proximity to fixed detection devices is not necessary. In addition, on-chip detection may be particularly useful when the chip is packaged in a housing, as is the case in certain embodiments of the invention.

**[0145]** III. Magnetic Particles

**[0146]** As will be evident, the magnetic particles to be coupled to the magnetic chip may be in any suitable form, including beads. For descriptive purposes the magnetic particles will be referred to herein as beads or magnetic beads, without thereby imposing any limitation on the size or shape of the particles. The beads may have any suitable size, depending upon the characteristics of the chip on which they are to be dispersed. In certain embodiments of the invention the beads are substantially spherical. For example, spherical beads with a diameter between about 1 and 10 microns may be used. In certain embodiments of the invention spherical beads with diameter between about 1 and 5 microns may be used. In certain embodiments of the invention spherical beads with a diameter between about 1 and 3 microns may be used. In addition, nanoparticles such as nanospheres may be used. The manufacturing technologies described above are compatible with fabrication of arrays with feature sizes down to the submicron scale, thus they can readily be employed to fabricate chips for use with nanoparticles.